

09H-2-DJ-D-B

<120> Recombinant Monoclonal Antibody Specific for Phosphotyrosine-Containing Proteins

<140>

<160> 12

<170> PatentIn Ver. 2.1

<210> 1

<211> 1365

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:cDNA for heavy chain of recombinant antibody

<400> 1

gagggtccagc	tgcarcagtc	tggacctgaa	ctggtgaagc	ctggggcttc	agtgatgata	60
tctctcagga	cttctgcata	cacattcact	gaaaacaccg	tgcactgggt	gaagcagagc	120
catggagaga	gccttgagtg	gattggaggt	attaatcctt	actatggtgg	ttctatcttc	180
agcccgaagt	tcaagggcaa	ggccacattg	actgtagaca	agtctctcag	cacagcctac	240
atggagctcc	gcagcctgac	atctgaggat	tctgcagttc	attactgtgc	aagaagggct	300
ggggcg tact	actttgacta	ctggggccaa	ggcaccactc	tcacagtctc	ctcagccaaa	360
acaacacccc	catcagttta	tccactggcc	cctgggtgtg	gagatacaac	tggttcctcc	420
gtgactctgg	gatgcctggt	caagggctac	ttccctgagt	cagtgactgt	gacttggaac	480
tctggatccc	tgtccagcag	tgtgcacacc	ttcccagctc	tcttgcagtc	tggactctac	540
actatgagca	gctcagtgac	tgtcccctcc	agcacctggc	caagtcagac	cgtcacctgc	600
agcgttgctc	accagaccag	cagcaccacg	gtggacaaaa	aacttgagcc	cagcggggccc	660
attttcaaca	tcaacccctg	tcctccatgc	aaggagtgtc	acaaatgcc	agctcctaac	720
ctcgagggtg	gaccatccgt	cttcatcttc	cctccaaata	tcaaggatgt	actcatgata	780
tccctgacac	ccaaggtcac	gtgtgtggtg	gtggatgtga	gcgaggatga	cccagacgtc	840
cagatcagct	ggttttgtgaa	caacgtggaa	gtacacacag	ctcagacaca	aacctataga	900
gaggattaca	acagtactat	ccgggtggtc	agcacccctc	ccatccagca	ccaggactgg	960
atgagtggca	aggagttcaa	atgcaaggtc	aacaacaaag	acctcccatc	acctatcgag	1020
agaaccatct	caaaaattaa	agggctagtc	agagctccac	aagtatacat	cttgccgcca	1080
ccagcagagc	agttgtccag	gaaagatgtc	agtctcaact	gcctggtcgt	gggcttcaac	1140

cctggagaca tcagtgtgga gtggaccagc aatgggcata cagaggagaa ctacaaggac 1200
accgcaccag tcctggactc tgacggttct tacttcatat atagcaagct caatatgaaa 1260
acaagcaagt gggagaaaac agattccttc tcatgcaacg tgagacacga gggctctgaaa 1320
aattactacc tgaagaagac catctcccg tctccgggta aatga 1365

<210> 2

<211> 645

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:cDNA for light
chain of recombinant antibody

<400> 2

gaaaatgtgc tcaccagtc tccagcaatc atgtctgcat ctccagggga aaaggtcacc 60
atgacctgca gggccagctc aagtgttaagt tccagttact tgcactggta tcggcagaag 120
tcaggtgcct cccccaact ctggatttat agcacatcca acttggcttc tggagtcctt 180
gtctcgcttca gtggcagtggt gtctgggacc tcttactctc tcacaatcag cagtgtggag 240
gctgaagatg ctgccactta ttactgccag cagtacagtg gttaccggac gttcgggtgga 300
ggcaccaagc tggaaatcaa acgggctgat gctgcaccaa ctgtatccat cttcccacca 360
tccagtgagc agttaacatc tggaggtgcc tcagtcgtgt gcttcttgaa caacttctac 420
cccagagaca tcaatgtcaa gtggaagatt gatggcagtg aacgacaaaa tgggtgtcctg 480
aacagttgga ctgatcagga cagcaaagac agcacctaca gcatgagcag caccctcaca 540
ttgaccaagg acgagtatga acgacataac agctatacct gtgaggccac tcacaagaca 600
tcaacttcac ccacgtcaa gagcttcaac aggaatgagt gttag 645

<210> 3

<211> 1389

<212> DNA

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:cDNA for heavy
chain of recombinant antibody with 3'-histidine
tag sequence

<400> 3

gaggtccagc tgcarcagtc tggacctgaa ctggtgaagc ctggggcttc agtgatgata 60
tcctgcagga cttctgcata cacattcact gaaaacaccg tgcactgggt gaagcagagc 120
catggagaga gccttgagtg gattggaggt attaatacctt actatggtgg ttctatcttc 180
agcccgaagt tcaagggcaa ggccacattg actgtagaca agtcctccag cacagcctac 240
atggagctcc gcagcctgac atctgaggat tctgcagtctt attactgtgc aagaagggct 300
ggggcgctact actttgacta ctggggccaa ggcaccactc tcacagtctc ctcagccaaa 360
acaacacccc catcagtcta tccactggcc cctgggtgtg gagataaac tgggttcctc 420
gtgactctgg gatgcctgggt caagggtctac ttccctgagt cagtgactgt gacttggaac 480

tctggatccc tgtccagcag tgtgcacacc ttcccagctc tccctgcagtc tggactctac 540
actatgagca gctcagtgac tgccccctcc agcacctggc caagtcagac cgtcacctgc 600
agcggttgctc acccagccag cagcaccacg gtggacaaaa aacttgagcc cagcggggccc 660
atttcaacaa tcaaccctg tccctccatgc aaggagtgtc acaaatgccc agtcctaac 720
ctcagagggtg gaccatccgt ctccatcttc cctccaaata tcaaggatgt actcatgac 780
tccctgacac ccaaggtcac gtgtgtggtg gtggatgtga gcgaggatga ccagacgctc 840
cagatcagct gggtttgtgaa caacgtggaa gtacacacag ctacagacaca aacccataga 900
gaggattaca acagtactat ccgggtgggc agcacccctc ccatccagca ccaggactgg 960
atgagtggca aggagttcaa atgcaaggtc aacaacaaag acctcccatc acccatcgag 1020
agaaccatct caaaaattaa agggctagtc agagctccac aagtatacat cttgccgcca 1080
ccagcagagc agttgtccag gaaagatgtc agtctcactt gcctggtcgt gggcttcaac 1140
cctggagaca tcagtgtgga gtggaccagc aatgggcata cagaggagaa ctacaaggac 1200
accgcaccag tccctggactc tgacggttct tacttcatat atagcaagct caatatgaaa 1260
acaagcaagt gggagaaaac agattccttc tcatgcaacg tgagacacga gggctctgaaa 1320
aattactacc tgaagaagac catctcccgg tctccgggta aagggtggcca tcaccaccat 1380
caccattga 1389

<210> 4

<211> 454

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:Amino acid
sequence for heavy chain of recombinant antibody

<400> 4

Glu Val Gln Leu Gln Gln Ser Gly Pro Glu Leu Val Lys Pro Gly Ala

1

5

10

15

Ser Val Met Ile Ser Cys Arg Thr Ser Ala Tyr Thr Phe Thr Glu Asn

20

25

30

Thr Val His Trp Val Lys Gln Ser His Gly Glu Ser Leu Glu Trp Ile

35

40

45

Gly Gly Ile Asn Pro Tyr Tyr Gly Gly Ser Ile Phe Ser Pro Lys Phe

50

55

60

Lys Gly Lys Ala Thr Leu Thr Val Asp Lys Ser Ser Ser Thr Ala Tyr

65

70

75

80

Met Glu Leu Arg Ser Leu Thr Ser Glu Asp Ser Ala Val Tyr Tyr Cys

85

90

95

Ala Arg Arg Ala Gly Ala Tyr Tyr Phe Asp Tyr Trp Gly Gln Gly Thr

100

105

110

Thr Leu Thr Val Ser Ser Ala Lys Thr Thr Pro Pro Ser Val Tyr Pro
115 120 125

Leu Ala Pro Gly Cys Gly Asp Thr Thr Gly Ser Ser Val Thr Leu Gly
130 135 140

Cys Leu Val Lys Gly Tyr Phe Pro Glu Ser Val Thr Val Thr Trp Asn
145 150 155 160

Ser Gly Ser Leu Ser Ser Ser Val His Thr Phe Pro Ala Leu Leu Gln
165 170 175

Ser Gly Leu Tyr Thr Met Ser Ser Ser Val Thr Val Pro Ser Ser Thr
180 185 190

Trp Pro Ser Gln Thr Val Thr Cys Ser Val Ala His Pro Ala Ser Ser
195 200 205

Thr Thr Val Asp Lys Lys Leu Glu Pro Ser Gly Pro Ile Ser Thr Ile
210 215 220

Asn Pro Cys Pro Pro Cys Lys Glu Cys His Lys Cys Pro Ala Pro Asn
225 230 235 240

Leu Glu Gly Gly Pro Ser Val Phe Ile Phe Pro Pro Asn Ile Lys Asp
245 250 255

Val Leu Met Ile Ser Leu Thr Pro Lys Val Thr Cys Val Val Val Asp
260 265 270

Val Ser Glu Asp Asp Pro Asp Val Gln Ile Ser Trp Phe Val Asn Asn
275 280 285

Val Glu Val His Thr Ala Gln Thr Gln Thr His Arg Glu Asp Tyr Asn
290 295 300

Ser Thr Ile Arg Val Val Ser Thr Leu Pro Ile Gln His Gln Asp Trp
305 310 315 320

Met Ser Gly Lys Glu Phe Lys Cys Lys Val Asn Asn Lys Asp Leu Pro
325 330 335

Ser Pro Ile Glu Arg Thr Ile Ser Lys Ile Lys Gly Leu Val Arg Ala
340 345 350

Pro Gln Val Tyr Ile Leu Pro Pro Pro Ala Glu Gln Leu Ser Arg Lys
355 360 365

Asp Val Ser Leu Thr Cys Leu Val Val Gly Phe Asn Pro Gly Asp Ile
370 375 380

Ser Val Glu Trp Thr Ser Asn Gly His Thr Glu Glu Asn Tyr Lys Asp
385 390 395 400

Thr Ala Pro Val Leu Asp Ser Asp Gly Ser Tyr Phe Ile Tyr Ser Lys
405 410 415

Leu Asn Met Lys Thr Ser Lys Trp Glu Lys Thr Asp Ser Phe Ser Cys
420 425 430

Asn Val Arg His Glu Gly Leu Lys Asn Tyr Tyr Leu Lys Lys Thr Ile
435 440 445

Ser Arg Ser Pro Gly Lys
450

<210> 5

<211> 214

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:Amino acid
sequence for light chain of recombinant antibody

<400> 5

Glu Asn Val Leu Thr Gln Ser Pro Ala Ile Met Ser Ala Ser Pro Gly
1 5 10 15

Glu Lys Val Thr Met Thr Cys Arg Ala Ser Ser Ser Val Ser Ser Ser
20 25 30

Tyr Leu His Trp Tyr Arg Gln Lys Ser Gly Ala Ser Pro Lys Leu Trp
35 40 45

Ile Tyr Ser Thr Ser Asn Leu Ala Ser Gly Val Pro Ala Arg Phe Ser
50 55 60

Gly Ser Gly Ser Gly Thr Ser Tyr Ser Leu Thr Ile Ser Ser Val Glu
65 70 75 80

Ala Glu Asp Ala Ala Thr Tyr Tyr Cys Gln Gln Tyr Ser Gly Tyr Arg
85 90 95

Lys	Gly	Lys	Ala	Thr	Leu	Thr	Val	Asp	Lys	Ser	Ser	Ser	Thr	Ala	Tyr	65	70	75	80
Met	Glu	Leu	Arg	Ser	Leu	Thr	Ser	Glu	Asp	Ser	Ala	Val	Tyr	Tyr	Cys	85	90	95	
Ala	Arg	Arg	Ala	Gly	Ala	Tyr	Tyr	Phe	Asp	Tyr	Trp	Gly	Gln	Gly	Thr	100	105	110	
Thr	Leu	Thr	Val	Ser	Ser	Ala	Lys	Thr	Thr	Pro	Pro	Ser	Val	Tyr	Pro	115	120	125	
Leu	Ala	Pro	Gly	Cys	Gly	Asp	Thr	Thr	Gly	Ser	Ser	Val	Thr	Leu	Gly	130	135	140	
Cys	Leu	Val	Lys	Gly	Tyr	Phe	Pro	Glu	Ser	Val	Thr	Val	Thr	Trp	Asn	145	150	155	160
Ser	Gly	Ser	Leu	Ser	Ser	Ser	Val	His	Thr	Phe	Pro	Ala	Leu	Leu	Gln	165	170	175	
Ser	Gly	Leu	Tyr	Thr	Met	Ser	Ser	Ser	Val	Thr	Val	Pro	Ser	Ser	Thr	180	185	190	
Trp	Pro	Ser	Gln	Thr	Val	Thr	Cys	Ser	Val	Ala	His	Pro	Ala	Ser	Ser	195	200	205	
Thr	Thr	Val	Asp	Lys	Lys	Leu	Glu	Pro	Ser	Gly	Pro	Ile	Ser	Thr	Ile	210	215	220	
Asn	Pro	Cys	Pro	Pro	Cys	Lys	Glu	Cys	His	Lys	Cys	Pro	Ala	Pro	Asn	225	230	235	240
Leu	Glu	Gly	Gly	Pro	Ser	Val	Phe	Ile	Phe	Pro	Pro	Asn	Ile	Lys	Asp	245	250	255	
Val	Leu	Met	Ile	Ser	Leu	Thr	Pro	Lys	Val	Thr	Cys	Val	Val	Val	Asp	260	265	270	
Val	Ser	Glu	Asp	Asp	Pro	Asp	Val	Gln	Ile	Ser	Trp	Phe	Val	Asn	Asn	275	280	285	
Val	Glu	Val	His	Thr	Ala	Gln	Thr	Gln	Thr	His	Arg	Glu	Asp	Tyr	Asn	290	295	300	
Ser	Thr	Ile	Arg	Val	Val	Ser	Thr	Leu	Pro	Ile	Gln	His	Gln	Asp	Trp	305	310	315	320

Met Ser Gly Lys Glu Phe Lys Cys Lys Val Asn Asn Lys Asp Leu Pro
 325 330 335

Ser Pro Ile Glu Arg Thr Ile Ser Lys Ile Lys Gly Leu Val Arg Ala
 340 345 350

Pro Gln Val Tyr Ile Leu Pro Pro Pro Ala Glu Gln Leu Ser Arg Lys
 355 360 365

Asp Val Ser Leu Thr Cys Leu Val Val Gly Phe Asn Pro Gly Asp Ile
 370 375 380

Ser Val Glu Trp Thr Ser Asn Gly His Thr Glu Glu Asn Tyr Lys Asp
 385 390 395 400

Thr Ala Pro Val Leu Asp Ser Asp Gly Ser Tyr Phe Ile Tyr Ser Lys
 405 410 415

Leu Asn Met Lys Thr Ser Lys Trp Glu Lys Thr Asp Ser Phe Ser Cys
 420 425 430

Asn Val Arg His Glu Gly Leu Lys Asn Tyr Tyr Leu Lys Lys Thr Ile
 435 440 445

Ser Arg Ser Pro Gly Lys Gly Gly His His His His His His
 450 455 460

<210> 7
 <211> 80
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence:HC 5' coding
 strand primer RAPHC-5

<400> 7
 gccaccatgg aatggagttg gatatttctc tttctcctgt caggaactgc aggtgtccac 60
 tctgaggtcc agctgcarca 80

<210> 8
 <211> 80
 <212> DNA
 <213> Artificial Sequence

<400> 8

007060"552755-090100

gccacatgg attttctggt gcagattttc agcttcttgc taatcagtgc ctcagttgca 60
atgtccagag gagaaaatgt 80

<210> 9
<211> 24
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:HC 3'
non-coding strand primer

<400> 9
ctaagctcat ttacccggag accg 24

<210> 10
<211> 25
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:LC 3'
non-coding strand primer

<400> 10
ctcaggacct ttgtctctaa cactc 25

<210> 11
<211> 62
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence:HC His 5'
coding strand primer

<400> 11
ctccccgtct cccggtaaag gtggccatca ccaccatcac cattgagctt agaagggcaa 60
tt 62

<210> 12
<211> 62
<212> DNA
<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence:HC His 5'
non-coding strand primer

<400> 12

aattgccctt ctaagctcaa tggatgatggg ggtgatggcc acctttaccc ggagaccggg 60
ag 62